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**TOXICITY TESTING OF SOIL SAMPLES
FROM J-FIELD, ABERDEEN PROVING GROUND, MD**

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RESEARCH AND TECHNOLOGY DIRECTORATE

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13. ABSTRACT (Maximum 200 words) Soil samples from the toxic burning pits, an area adjacent to the toxic burning pits, white phosphorus pits, and riot control pits were tested for their toxicity to lettuce and earthworms as part of an ecological risk assessment of J-Field. To adequately assess the toxicity of chemicals in the soil, it was important to determine effects at several trophic levels. Therefore, standardized screening and definitive phytotoxicity and earthworm toxicity tests were used in the study. If a screening test produced lethal effects, then a definitive test was conducted. Definitive tests used concentrations of 25, 50, 75, and 100% (dry weight basis). Test results from the toxic burning pits showed the overall toxicity to be nontoxic at two sites (JBPPC and JBPMMA); minimally toxic at four sites (JBP1C, JBP2C, JBPPA, and JBPNP); moderately toxic at two sites (JBPPB and JBPMC); and highly toxic at one site (JHDP). An additional site (JBPMB) was not fully evaluated. Results from the white phosphorus pits were nontoxic for three sites (JWP1E, JWPPB, and JWP2C) and moderately toxic at one site (JWPPA). One site (JBT1W) from the riot control pits produced minimal toxicity. The majority of the toxicological effects were exhibited by earthworms. Testing with bioassays containing different trophic levels identified toxic sites and sites/areas needing more investigation.				
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PREFACE

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TOXICITY TESTING OF SOIL SAMPLES FROM J-FIELD, ABERDEEN PROVING GROUND, MD

1. INTRODUCTION

Open burning was the standard practice for disposing of toxic chemicals at Aberdeen Proving Ground, MD, from the late 1940's through the 1960's. Typically, open trenches or pits were dug, filled with wood, and soaked with fuel oil. Containers filled with materials to be disposed were placed on top of the wood, and the fuel was ignited. The containers were opened with an explosive charge, which allowed the materials to be incinerated in place. This disposal site (pits and surrounding area), consisting of 135 acres of woodlands, grasslands, and marshlands (bordered on the east by the Bush River and on the west by the Gunpowder Rivers; all tributaries of the Chesapeake Bay) was known as the J-Field disposal site. The site was used to dispose of a variety of U.S. Army-unique chemicals, laboratory waste, pilot plant operations, and high explosive rounds through burning or detonation. The area consisted of several types of disposal pits (e.g., toxic burning, white phosphorus, and riot control pits), ranging in size from 15 by 300 ft² (41.8 m²) to 4 by 6 ft² (2.2 m²). This site has since been listed on the National Priority List (NPL) for site clean up by the U.S. Environmental Protection Agency (EPA).

Determining the environmental toxicity of disposed materials at this site is important in developing an ecological risk assessment. Toxicity tests have been used successfully in risk assessments at other terrestrial sites.^{1,2} To adequately assess the chemical toxicity to the terrestrial community, it is important to determine effects at several trophic levels. Plants (seed germination and early seedling growth test) and earthworms (survival and growth rates), representing two trophic levels, were chosen for this study.

The use of plants and earthworms as measurement endpoints was done for several reasons. One reason is that chemicals may adversely damage the ecosystem and negatively impact wildlife that feed on plants (e.g., deer) and earthworms (e.g., birds). In addition, earthworms are considered key organisms in the soil community. They increase the fertility of soil by increasing the availability of nutrients, and they are also an important link in the food chain. Earthworms are important to the terrestrial ecosystem; therefore, their use in assessing the chemical hazards to the ecosystem is important.

The role of the U.S. Army Edgewood Research, Development and Engineering Center's (ERDEC) Environmental Technology Team was to determine baseline environmental toxicity data on the soils from various pits located at the site. This study was part of a joint effort between Argonne National Laboratory (ANL, Argonne, IL) and ERDEC personnel. Members of ANL provided ERDEC personnel with soil samples from the toxic burning pits, an area adjacent to the toxic burning pits, the white phosphorus pits, and the riot control pits. ERDEC personnel were responsible for the terrestrial toxicity data for this site. This report presents the results of the ERDEC program.

2. MATERIALS AND METHODS

2.1 Soil Sampling Sites.

Soil samples were collected by ANL personnel from four locations at J-Field. The location and number of samples collected from each were as follows: (1) toxic burning pits (10 samples); (2) white phosphorus pits (4 samples); (3) riot control pits (2 samples); and areas adjacent to the toxic burning pits (2 samples).

2.2 Preparation of Soil Samples.

Soil samples were sieved through a 5 mm² mesh wire screen to remove rocks and stones, twigs, leaves, and other large debris. After sieving, each soil was placed back into its original plastic sampling bag to retain soil moisture. Dry-fraction and water-holding capacity determinations were made for each sample to determine the quantity of water required to bring the soil up to nominal field moisture levels before phytotoxicity and earthworm toxicity tests were conducted.

2.3 Dry Fraction Determination.

The dry fraction of each soil sample was determined by placing a 2-3 g subsample of soil into a weighed aluminum weighing pan. After a total weight was obtained, the pans were placed into a drying oven (110 °C) for 3 days. At the end of this time, the samples were weighed again to obtain a soil dry weight. The dry weight of soil divided by the initial weight of soil yielded the dry fraction.

2.4 Water-Holding Capacity Determination.

Subsamples from each representative soil type were used to determine the water-holding capacity. This was accomplished by placing a known amount of soil (approximately 10 g) into a 25 x 45 mm² polyethylene column sealed with silicone sealant on a porous ceramic plate so that the soil made direct contact with the plate. The soils in the columns were wetted with distilled water and allowed to settle for 4 hr. Additional water (10 mL) was then added to each column. A partial negative pressure of 0.3 atmospheres (30-35 KPa) was applied to the soils via the porous ceramic plate, and the columns remained under tension for 24 hr. At the end of this time, the soil from each column was removed and reweighed. The difference between the initial soil weight and the final soil weight was used to determine water-holding capacity (WHC) using the following formula:

$$\text{WHC} = \frac{(100\%) [(\text{final wt}) - (\text{dry fraction}) \times (\text{initial wt})]}{(\text{dry fraction}) \times (\text{initial wt})}$$

2.5 Toxicity Testing.

2.5.1 Phytotoxicity Tests.

The testing regimen for determining phytotoxicity was adapted from the EPA's Early Seedling Growth Toxicity Test³ (Toxicology Division SOP Nos. LTP-62 - 65 and Research Protocol No. 22093000X059). The testing regimen used for all of the plant tests (screening and definitive) is summarized below.

For each soil sample tested, approximately 800 g of pea gravel was placed into a 150-mm (diameter) flower pot. A single layer of cheesecloth was placed on top of the gravel, and 800 g (dry weight) of field-moist soil was added to the pot to bring the soil level to within 1 cm of the top.

Positive and negative controls were incorporated into the experimental design using soil obtained from an area along Winters Run (WR). This soil was used as the reference soil (for the plant and earthworm tests) since an appropriate soil was not found at J-Field. The physical and chemical properties of WR soil are provided in Table 1.

Table 1. Physical and Chemical Characteristics of WR Soil

Soil Parameters*		
Mechanical Analysis	Soil Analysis	
% sand 49	NO ₃ (Lb/A)	1.9
	P ₂ O ₅ * (Lb/A)	20.0
	K ₂ O* (Lb/A)	185.0
% silt 37	Ca (Lb/A)	1328.0
	Mg (Lb/A)	203.0
	Mn (Lb/A)	331.0
% clay 14	Zn (Lb/A)	4.8
	Cu (Lb/A)	5.0
	CEC (meq/100 g)	7.6
Texture - loam	pH	5.7
	% organic matter	4.4

*Determined by the Soil Testing Laboratory, University of Maryland, College Park, MD; P expressed as P₂O₅; K expressed as K₂O

The screening test consisted of three replicates of unaltered J-Field soil and appropriate positive and negative controls. The definitive test consisted of mixing a subsample of the J-Field soil with a quantity of reference soil (WR; dry weight basis)

to produce sample soil concentrations of 25, 50, 75, and 100%. The definitive test was also performed in triplicate with appropriate positive (WR+) and negative (WR-) controls.

The WR+ control used in plant testing used a spike of copper sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) at a target concentration of 1,000 μg Cu/g soil. The WR- control was WR soil without the spike added.

The plant species used in the screening test was lettuce (*Lactuca sativa*, L; var. Black Seeded Simpson; Meyer Seed Company, Baltimore, MD; lot B1-394). Lettuce seeds were sorted to remove broken or malformed seeds and to obtain seeds of similar size. Twenty seeds were planted per pot for each soil sample. Pots were watered to WHC. After emergence, the seedlings were thinned to the 10 most uniform per pot. "Day 1" of treatment was determined when 50% of the total number of control seeds had emerged. A record of the rate of seed emergence was made over the 14-day study period as a direct measure of effective seed germination. Plant height measurements were taken 4 times during the study period. Any plant abnormalities (e.g., chlorosis, necrosis, etc.) were noted and reported in results and discussion. A final measurement was made when plants were harvested (Day 14). After harvest, plant fresh and dry weight measurements were made as additional measures of plant growth.

Data were produced on the seed emergence rates, plant heights, and plant dry weights. Statistical evaluations of plant data included Analysis of Variance (ANOVA) and Student-Newman-Keuls multiple range tests.⁴

2.5.2 Earthworm Toxicity Tests.

Earthworm toxicity screening tests used the earthworm (*Eisenia foetida*) in the screening and definitive tests. Survival rates and differences between initial and final weights (sublethal endpoints) were used as indices of toxicity.

The test methods used for earthworm toxicity studies were adapted from Karnak and Hamelink⁵ and Neuhauser *et al.*⁶ The screening regimen for determining earthworm toxicity (Toxicology Division SOP No. LTP-48 and Research Protocol No. 22093000X059) is summarized below.

Earthworms, originally purchased from Bert's Bait Farm (Irvine, KY), were bred and housed in styrofoam coolers in our laboratory. Earthworms were housed under controlled temperature in a low-temperature incubator (21.0 ± 0.2 °C) during the course of the studies. An earthworm toxicity screening test consisted of placing a set of five earthworms into each of three 600-mL glass beakers per soil sample (i.e., three replicates per sampling location).

For each replicate, 200 g of unaltered J-Field soil (dry weight) was mixed in a food blender with a sufficient quantity of distilled water to bring the soil moisture level up to nominal field capacity. This was mixed for approximately 3 min until uniformly mixed and then placed into one of the beakers. The procedure was then repeated for the other beakers, and included WR+ and WR- controls. The WR+ control had a spike of paranitrophenol added at a target concentration of either 30 or 50 $\mu\text{g/g}$.

After the beakers were prepared, 75-100 earthworms were removed from one of the styrofoam coolers and put into a plastic container. The earthworms were quickly rinsed in tap water, and excess water drained from the pan. Five earthworms were arbitrarily selected, quickly blotted with a paper towel, and weighed as a group. They were then placed into one of the beakers. After five earthworms had been added to each beaker, the beakers were covered with nylon screen and cheesecloth held in place by a rubber band. They were then placed in plastic trays within the incubator. Water was added to the trays to help prevent the soil in the beakers from drying out. The incubator lights were set for continuous operation. Since the earthworms are photophobic, the light encouraged them to burrow into the soil being tested and helped prevent them from crawling out of the beakers.

The earthworms were housed in the incubator for the 14-day exposure period. Beakers were re-randomized in the trays on Day 7. On Day 14, the earthworms were removed from each beaker and re-weighed to obtain a final weight; they were also examined for their physical condition; any changes that occurred in physical condition (e.g., color, texture, motility, etc.) were noted and reported in Sections 3 and 4.

A definitive test consisted of mixing a subsample of a soil sample with a quantity of reference soil (WR; dry weight basis) to produce J-Field soil concentrations of 25, 50, 75, and 100%. The definitive test was also performed in triplicate with appropriate WR+ and WR- controls.

Results from the screening and definitive tests produced data on initial and final weights, survivorship rates, and weight differences. Two statistical methods were used to evaluate earthworm definitive test data: the Analysis of Covariance (ANCOVA) (to test weight differences), and the T-test pairwise comparison of least square means (means adjusted for the covariate).⁴

3. RESULTS

Data from phytotoxicity screening and definitive tests were summarized by location. Indices of toxicity for the screening tests included the seed emergence rate (SER), average plant height, and average plant dry weight for each soil sample tested. An ANOVA on plant heights and dry weights was performed on data from the definitive tests.

Earthworm toxicity data included an earthworm survival rate (ESR) (%) and differences between initial and final weights. An ANCOVA and T-Test analysis were performed on definitive test data. These results were also summarized by location.

Data and statistical information for all of the bioassays are given in Appendixes A through D.

3.1 Toxic Burning Pits.

The results of the phytotoxicity screening tests using lettuce seeds are presented in Table 2. The JBP2C and JBP1C soils had SERs of 100%. The JBPMB soil had an SER of 100%; but, none of the seedlings survived to Day 14. Some soils

(JBPPC, JBPMMA, and JBPPA) had SERs of 75%. Although JBPPC soil had an SER value of only 75%, it had one of the largest average plant heights and one of the greatest average dry weights of all phytotoxicity tests. Although the JBPPB soil had an SER of only 60%, it had the largest average plant height but a relatively small average dry weight compared to the other soil samples. Because an insufficient quantity of JBPMC was provided, no phytotoxicity screening test was conducted on this sample. None of the seeds planted in JHDP soil germinated.

Table 2. Results of Phytotoxicity Screening of Soils from Toxic Burning Pits (20 seeds/soil sample) - Day 14

Soil ID	SER (%)	Surviving Plants (10 max)	Avg Plant Ht (mm)	Avg Dry Wt (g)
JBP2C	100	10	8.5	0.0071
JBP1C	100	10	7.2	0.0046
JBPMB	100	0	0.0	0.0000
JBPPC	75	10	10.8	0.0065
JBPMMA	75	10	8.9	0.0053
JBPPA	75	10	8.8	0.0051
JBPPB	60	10	10.9	0.0033
JBPMC	NA	NA	NA	NA
JHDP	0	0	0.0	0.0000
WR-	65	10	8.1	0.0061
WR+	25	3	2.0	0.0007

Since none of the seeds planted in JHDP soil germinated during the screening test, a definitive phytotoxicity test was conducted to determine JHDP's toxicity on lettuce (Table 3). At the 100% level, there was an SER of 75%. The SER dropped to 47% at the 75% level and remained at similar values at the 25 and 50% levels. The WR- control had a 38% SER, and the WR+ control had only a 15% SER. The average plant heights of lettuce grown in JHDP soil at the 75 and 100% levels were less than those at the lower levels (i.e., 25, 50, and WR-). An ANOVA of the plant heights showed a significant ($p < 0.0001$) difference between treatments. The Student-Newman-Keuls (SNK) analysis placed the WR-, 25, and 50% levels in one group and the WR+, 75, and 100% levels in another group. The average dry weights at the 75 and 100% levels were also less than those at the lower levels, but the ANOVA and SNK of the dry weights showed that this difference was insignificant.

Table 3. Results of Phytotoxicity Testing of JHDP Soil from Toxic Burning Pits (60 seeds/treatment) - Day 14

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	38	23	7.96 ± 2.14	0.0069 ± 0.0012
25%	53	27	7.67 ± 2.09	0.0079 ± 0.0008
50%	55	25	7.68 ± 1.75	0.0087 ± 0.0026
75%	47	25	5.00 ± 1.66	0.0067 ± 0.0019
100%	75	27	5.37 ± 1.42	0.0064 ± 0.0012
WR+	15	9	5.56 ± 1.94	0.0023 ± 0.0007

An additional soil sample, JBPNP, was received after the initial screening tests were conducted. A definitive test (Table 4) was conducted to determine the toxicity of this soil on lettuce. The SERs were 95% or greater at all levels except for WR+ (23%). Average plant heights were smaller at the 75 and 100% levels (ANOVA was not significant at $p > 0.05$), but average dry weights were greater at the same levels (ANOVA was significant at $p < 0.0004$). The overall indication was that no lethal or sublethal effects were produced.

Table 4. Results of Phytotoxicity Testing of JBPNP Soil from Toxic Burning Pits (60 seeds/treatment) - Day 14

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	95	29	14.55 ± 2.75	0.0069 ± 0.0006
25%	95	30	14.30 ± 2.25	0.0085 ± 0.0017
50%	98	30	14.23 ± 1.99	0.0098 ± 0.0012
75%	97	30	13.73 ± 1.76	0.0110 ± 0.0004
100%	95	30	13.80 ± 1.99	0.0123 ± 0.0013
WR+	23	14	3.36 ± 1.08	0.0007 ± 0.0006

The results of the earthworm screening tests (Table 5) indicated lethal and sublethal effects at JBPMB, JBPPB, JBPMC, and JHDP locations. However, the toxic effects of JBPMB, JBPPB, and JBPMC soils were minimal when compared to the effects of JHDP soil on earthworms. In the JHDP soil, the surviving earthworms were found clinging to the sides of the beakers above the soil line, thus surviving the 14-day study by not penetrating the soil. The ESR was 93-100% for all other soil samples.

Table 5. Results of Earthworm Toxicity Screening of Soils from Toxic Burning Pits

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (\pm)	Avg Wt Diff/ Sample	Effect
JBP2C	A	100	0.35	0.31	- 0.04	- 0.07	sublethal
	B		0.39	0.30	- 0.09		
	C		0.42	0.33	- 0.09		
JBP1C	A	100	0.46	0.40	- 0.06	- 0.10	sublethal
	B		0.53	0.42	- 0.11		
	C		0.54	0.42	- 0.12		
JBPMB	A	93	0.38	0.25	- 0.13	- 0.12	lethal/ sublethal
	B		0.43	0.38	- 0.05		
	C		0.60	0.41	- 0.19		
JBPPC	A	100	0.45	0.47	+ 0.02	+ 0.003	none
	B		0.30	0.30	0.00		
	C		0.39	0.38	- 0.01		
JBPMA	A	100	0.41	0.40	- 0.01	- 0.02	none
	B		0.35	0.29	- 0.06		
	C		0.34	0.35	+ 0.01		
JBPPA	A	100	0.50	0.37	- 0.13	- 0.12	sublethal
	B		0.48	0.44	- 0.04		
	C		0.49	0.30	- 0.19		
JBPPB	A	93	0.66	0.49	- 0.17	- 0.10	lethal/ sublethal
	B		0.47	0.42	- 0.05		
	C		0.48	0.41	- 0.07		
JBPMC	A	93	0.49	0.35	- 0.14	- 0.08	lethal/ sublethal
	B		0.51	0.47	- 0.04		
	C		0.55	0.50	- 0.05		
JHDP	A	40	0.41	none	NA	- 0.15	lethal/ sublethal
	B		0.35	0.27	- 0.08		
	C		0.47	0.26	- 0.21		
WR-	A	100	0.44	0.45	+ 0.01	+ 0.04	none
	B		0.50	0.59	+ 0.09		
	C		0.43	0.44	+ 0.01		
WR+ (50)	A	0	0.39	none	NA	NA	lethal
	B		0.42	none	NA		
	C		0.42	none	NA		

A definitive earthworm toxicity test (Table 6) was conducted on JHDP soil because it had produced lethal and sublethal effects in both the seed emergence and earthworm screening tests. This soil was mixed with an appropriate amount of WR soil (JHDP/WR; dry weight basis) to produce concentrations of 25, 50, 75, and 100%. Lethal and sublethal effects were produced at the 75 and 100% levels. Overall, the lethal and sublethal effects diminished as the amount of JHDP soil in the mixture decreased. At the WR- and 25% levels, the earthworms averaged a slight weight gain.

The ANCOVA of earthworm weights showed that the difference between the initial and final weights was very highly significant ($p < 0.0001$) among treatment levels. The results of the T-test indicated a highly significant ($p < 0.01$) difference between the 100% level and all other levels. There was also a significant ($p < 0.05$) difference between the WR- and 25% levels and the 50, 75, and 100% levels. However, there was no significant ($p > 0.05$) difference between the WR- and the 25% levels.

Table 6. Results of Earthworm Toxicity Testing of JHDP Soil

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (\pm)	Avg Wt Diff/Sample	Effect
WR-	A	100	0.40	0.40	0.00	+ 0.01	none
	B		0.39	0.39	0.00		
	C		0.60	0.63	+ 0.03		
25%	A	100	0.48	0.46	- 0.02	+ 0.01	none
	B		0.36	0.39	+ 0.03		
	C		0.46	0.48	+ 0.02		
50%	A	100	0.34	0.30	- 0.04	- 0.06	sublethal
	B		0.43	0.34	- 0.09		
	C		0.43	0.37	- 0.06		
75%	A	93	0.39	0.31	- 0.08	- 0.08	lethal/ sublethal
	B		0.37	0.27	- 0.10		
	C		0.39	0.34	- 0.05		
100%	A	47	0.40	0.24	- 0.16	- 0.19	lethal/ sublethal
	B		0.58	0.43	- 0.15		
	C		0.61	0.35	- 0.26		
WR + (50)	A	0	0.40	none	NA	NA	lethal
	B		0.42	none	NA		
	C		0.43	none	NA		

An additional earthworm screening test (Table 7) was conducted on a soil sample, JBPNP, which was received after the initial screening tests were conducted. The ESR was 100%, thus indicating no lethal effects. A greater weight loss at the 100% JBPNP level indicated that some sublethal effects may have occurred.

Table 7. Results of Earthworm Toxicity Screening of JBPNP Soil from Toxic Burning Pits

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (\pm)	Avg Wt Diff/ Sample	Effect
JBPNP	A	100	0.48	0.38	- 0.10	- 0.11	sublethal
	B		0.51	0.40	- 0.11		
	C		0.54	0.42	- 0.12		
WR-	A	100	0.57	0.55	- 0.02	- 0.02	none
	B		0.44	0.42	- 0.02		
	C		0.49	0.46	- 0.03		
WR+ (30)	A	0	0.53	none	NA	NA	lethal
	B		0.58	none	NA		
	C		0.54	none	NA		

3.2 White Phosphorus Pits.

The results of the phytotoxicity screening test (Table 8) indicated no lethal or sublethal effects on lettuce seeds since the SERs for all soils from this location ranged from 70-95%, all test plants survived, plant heights looked good, and dry weights were adequate although the dry weight of JWPPA was somewhat low.

The results of the earthworm screening test (Table 9) indicated no lethal or sublethal effects of these soils (except in JWPPA soil) on earthworms. The effect was a small decline in the ESR and an average weight loss slightly greater than the other samples.

3.3 Riot Control Pits.

The results of the phytotoxicity screening tests using lettuce are presented in Table 10. Although JBT1W had only a 40% emergence rate, the average plant height and average dry weight were greater than or equal to the WR- controls. No seedlings emerged in JBTMA soil.

Table 8. Results of Phytotoxicity Screening of Soils from White Phosphorus Pits (20 seeds/soil sample) - Day 14

Soil ID	SER (%)	Surviving Plants (10 max)	Avg Plant Ht (mm)	Avg Dry Wt (g)
JWP1E	95	10	9.5	0.0143
JWPPB	80	10	11.8	0.0055
JWP2C	70	10	10.2	0.0123
JWPPA	70	10	11.6	0.0029
WR-	65	10	8.1	0.0061
WR+	25	3	2.0	0.0007

Table 9. Results of Earthworm Toxicity Screening of Soils from White Phosphorus Pits

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (\pm)	Avg Wt Diff/Sample	Effect
JWP1E	A	100	0.43	0.42	- 0.01	- 0.02	none
	B		0.43	0.42	- 0.01		
	C		0.40	0.37	- 0.03		
JWPPB	A	100	0.42	0.45	+ 0.03	+ 0.02	none
	B		0.55	0.57	+ 0.02		
	C		0.50	0.52	+ 0.02		
JWP2C	A	100	0.55	0.54	- 0.01	+ 0.01	none
	B		0.49	0.54	+ 0.05		
	C		0.52	0.52	0.00		
JWPPA	A	93	0.41	0.40	- 0.01	- 0.05	lethal/ sublethal
	B		0.50	0.43	- 0.07		
	C		0.47	0.39	- 0.08		
WR-	A	100	0.44	0.45	+ 0.01	+ 0.04	none
	B		0.50	0.59	+ 0.09		
	C		0.43	0.44	+ 0.01		
WR+ (50)	A	0	0.39	none	NA	NA	lethal
	B		0.42	none	NA		
	C		0.42	none	NA		

Table 10. Results of Phytotoxicity Screening of Soils from Riot Control Pits (20 seeds/soil sample) - Day 14

Soil ID	SER (%)	Surviving Plants (10 max)	Avg Plant Ht (mm)	Avg Dry Wt (g)
JBT1W	40	8	11.25	0.0061
JBTMA	0	0	NA	NA
WR-	65	10	8.1	0.0061
WR+	25	3	2.0	0.0007

Since the SERs were low for the JBT1W and JBTMA soils, a definitive phytotoxicity test was conducted for each one. The results for JBT1W soil are provided in Table 11. The SERs ranged from 55 to 83% for lettuce seeds grown in this soil. There was no significant (ANOVA: $p > 0.2763$) difference in plant heights between the different treatment levels. Similar results were found for the dry weights (ANOVA: $p > 0.2727$).

Table 11. Results of Phytotoxicity Testing of JBT1W Soil from Riot Control Pits (60 seeds/treatment) - Day 14

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	62	30	9.57 ± 2.34	0.0065 ± 0.0016
25%	82	30	10.50 ± 2.42	0.0080 ± 0.0006
50%	83	30	9.63 ± 1.88	0.0076 ± 0.0009
75%	62	26	10.69 ± 3.38	0.0070 ± 0.0010
100%	55	26	10.31 ± 1.78	0.0059 ± 0.0015
WR+	0	0	0.00 ± 0.00	0.0000 ± 0.0000

The results for JBTMA soil are provided in Table 12. The SERs ranged from 93 to 100%, except for WR+ (38%). The ANOVA and SNK of plant heights indicated a significant ($p < 0.0001$) difference between WR+ and WR- and the other treatment levels. Similar results were found for the dry weights.

The results of the earthworm screening test (Table 13) indicate no lethal effects on earthworms from the JBT1W and JBTMA soil samples. In the JBT1W soil, results indicated a sublethal effect (average weight loss) on earthworms.

Table 12. Results of Phytotoxicity Testing of JBTMA Soil from Riot Control Pits (60 seeds/treatment) - Day 15

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	93	30	16.47 ± 2.54	0.0073 ± 0.0008
25%	98	30	13.33 ± 2.87	0.0049 ± 0.0003
50%	100	30	13.07 ± 2.24	0.0045 ± 0.0004
75%	100	30	11.40 ± 2.01	0.0045 ± 0.0006
100%	98	30	10.67 ± 2.99	0.0045 ± 0.0009
WR+	38	23	5.39 ± 1.80	0.0013 ± 0.0003

Table 13. Results of Earthworm Toxicity Screening of Soils from Riot Control Pits

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (±)	Avg Wt Diff/Sample	Effect
JBT1W	A	100	0.53	0.53	0.00	- 0.03	sublethal
	B		0.50	0.49	- 0.01		
	C		0.53	0.46	- 0.07		
JBTMA	A	100	0.56	0.66	+ 0.10	+ 0.04	none
	B		0.53	0.57	+ 0.04		
	C		0.59	0.58	- 0.01		
WR-	A	100	0.44	0.45	+ 0.01	+ 0.04	none
	B		0.50	0.59	+ 0.09		
	C		0.43	0.44	+ 0.01		
WR+ (50)	A	0	0.39	none	NA	NA	lethal
	B		0.42	none	NA		
	C		0.42	none	NA		

3.4 Adjacent Areas.

In the phytotoxicity screening tests (Table 14), JBPGF soil yielded a 10% SER, with only 2 seedlings surviving to Day 14. The JBPCP104 soil produced a 30% seed emergence rate with 5 seedlings surviving to Day 14.

Table 14. Results of Phytotoxicity Screening of Soils from Adjacent Areas (20 seeds/soil sample) - Day 14

Soil ID	SER (%)	Surviving Plants (10 max)	Avg Plant Ht (mm)	Avg Dry Wt (g)
JBPGF	10	2	6.0	0.0029
JBPCP104	30	5	4.6	0.0035
WR-	65	10	8.1	0.0061
WR+	25	3	2.0	0.0007

Since the SERs and seedling survivorship rates were low for the JBPGF and JBPCP104 soils, a definitive phytotoxicity test was conducted for each one. The results are given in Table 15 for JBPGF soil. The SERs ranged from 82 to 100% for lettuce seeds grown in this soil.

The ANOVA of plant heights indicated a significant ($p < 0.0001$) difference between WR- and the other levels. However, the ANOVA of dry weights presented no significant ($p > 0.05$) difference in and among the WR- and JBPGF treatments.

Table 15. Results of Phytotoxicity Testing of JBPGF Soil from Adjacent Areas (60 seeds/treatment) - Day 15

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	48	26	10.27 \pm 3.39	0.0033 \pm 0.0015
25%	82	30	7.87 \pm 2.18	0.0053 \pm 0.0017
50%	93	30	8.00 \pm 1.44	0.0046 \pm 0.0004
75%	100	30	7.97 \pm 1.16	0.0038 \pm 0.0002
100%	92	30	7.20 \pm 1.54	0.0060 \pm 0.0004
WR+	0	0	0.00 \pm 0.00	0.0000 \pm 0.0000

Definitive phytotoxicity testing of JBPCP104 soil (Table 16) indicated no lethal effects on lettuce although the SERs were lower than those of some other soils tested. The ANOVA and SNK of plant heights indicated that the only significant ($p < 0.0001$) difference was between WR+ and the other treatments. However, the ANOVA and SNK for the dry weights showed a significant ($p < 0.0001$) difference between WR+ and WR- and the other treatment levels.

Table 16. Results of Phytotoxicity Testing of JBPCP104 Soil from Adjacent Areas (60 seeds/treatment) - Day 14

Soil ID	SER (%)	Surviving Plants (30 max)	Avg Plant Ht (mm) + Std Dev	Avg Dry Wt (g) + Std Dev
WR-	42	25	10.28 ± 3.27	0.0043 ± 0.0006
25%	77	30	10.03 ± 2.14	0.0097 ± 0.0006
50%	62	30	10.37 ± 2.09	0.0107 ± 0.0021
75%	63	29	8.59 ± 2.23	0.0103 ± 0.0012
100%	63	30	8.97 ± 1.47	0.0083 ± 0.0006
WR+	10	5	5.40 ± 1.14	0.0012 ± 0.0002

The results of the earthworm screening test (Table 17) indicated no negative impact of JBPGF soil on earthworms; however, lethal and sublethal effects on earthworms were found for JBPCP104 soil. Since the earthworm screening indicated potential for lethal/sublethal effects for JBPCP104, a definitive earthworm test was conducted. The results (Table 18) indicated this soil produced no lethal effects on earthworms. The ANCOVA showed a highly significant ($p < 0.0016$) difference in average earthworm weights between the 25% and other levels, except the 50% one. The T-Test analysis indicated a significant ($p < 0.05$) difference between the 25 and 50% and the other levels. Earthworms in the 25 and 50% levels were the only ones that had overall weight gains. Weight differences of earthworms in the 75 and 100% levels were not significantly ($p < 0.05$) different from the controls. However, they did have weight losses significantly ($p \leq 0.0133$) different than those in the 25 and 50% levels, indicating possible sublethal effects.

4. DISCUSSION

Data from the phytotoxicity and earthworm toxicity screening tests were used to determine which soils merited further testing. Phytotoxicity testing of soils from the toxic burning pits indicated some toxic effects on lettuce seed germination and growth. Samples JBPMB, JBPPB, and JHDP were the soils producing the most toxic effects on lettuce.

Although JBPMB had an SER of 100%, none of the seedlings survived to the end of the 14-day study. The earthworm toxicity screening test of this soil produced an ESR of 93% (one of the 15 earthworms died), and a sublethal effect was noted since the average weight difference was -0.12 g. Definitive phytotoxicity or earthworm toxicity studies of this soil were not conducted. Additional studies of this soil are recommended to determine the severity of toxicity.

Table 17. Results of Earthworm Toxicity Screening of Soils from Adjacent Areas

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (±)	Avg Wt Diff/Sample	Effect
JBPGF	A	100	0.39	0.43	+ 0.04	+ 0.04	none
	B		0.45	0.50	+ 0.05		
	C		0.57	0.60	+ 0.03		
JBPCP-104	A	80	0.51	0.34	- 0.17	- 0.13	lethal/ sublethal
	B		0.45	0.35	- 0.10		
	C		0.33	0.22	- 0.11		
WR-	A	93	0.34	0.34	0.00	+ 0.003	none
	B		0.47	0.49	+ 0.02		
	C		0.43	0.42	- 0.01		
WR + (50)	A	0	0.40	none	NA	NA	lethal
	B		0.42	none	NA		
	C		0.43	none	NA		

Phytotoxicity screening of JBPPB soil produced an SER of 60% with a low average dry weight. The earthworm toxicity screening test also produced some lethal and sublethal effects. Because ANL personnel could not relocate the exact sampling site to obtain additional soil samples with which to conduct a definitive study, JBPNP soil was substituted for the JBPPB soil. A definitive phytotoxicity test showed the JBPNP soil to have neither lethal nor sublethal effects on seed germination or seedling growth. The earthworm toxicity screening test of JBPNP soil produced no lethal effect (ESR = 100%) and only a sublethal effect with an average weight loss of -0.11 g. Based on the results of the screening tests, no definitive tests were conducted on the JBPNP soil, and additional JBPPB soil testing is recommended if its exact location can be redetermined. Such action will enable investigators to define the severity of its toxicity better. The JBPNP soil did not appear to be an effective substitute for JBPPB soil, since the JBPNP caused no phytotoxicity and only minor sublethal negative effects on earthworms.

Phytotoxicity screening of JHDP soil produced a 0% SER. The definitive phytotoxicity test showed significantly decreased plant heights at the higher concentrations (75 and 100%) than the lower concentrations with ANOVA significant at $p < 0.0001$. However, the ANOVA and SNK of plant dry weights indicated no significant ($p > 0.05$) difference. The earthworm toxicity screening test of JHDP soil produced a 40% ESR with an average weight loss of -0.15 g. Therefore, a definitive test was conducted. The ANCOVA indicated a significant ($p < 0.0001$) difference between treatments. The T-Test analysis showed a significant difference between the 100% and all other levels except 75%. Earthworms that survived at the 100% level

Table 18. Results of Earthworm Toxicity Testing of JBPCP104 Soil

Soil ID	Rep	ESR (%)	Initial Avg Wt (g)	Final Avg Wt (g)	Wt Diff (\pm)	Avg Wt Diff/Sample	Effect
WR-	A	100	0.69	0.61	- 0.08	- 0.04	none
	B		0.58	0.55	- 0.03		
	C		0.57	0.57	0.00		
25%	A	100	0.58	0.61	+ 0.03	+ 0.04	none
	B		0.55	0.60	+ 0.05		
	C		0.62	0.65	+ 0.03		
50%	A	93	0.54	0.58	+ 0.04	+ 0.02	none
	B		0.61	0.64	+ 0.03		
	C		0.55	0.54	- 0.01		
75%	A	100	0.59	0.55	- 0.04	- 0.05	none
	B		0.50	0.47	- 0.03		
	C		0.68	0.61	- 0.07		
100%	A	100	0.61	0.56	- 0.05	- 0.07	possible sublethal effect
	B		0.62	0.56	- 0.06		
	C		0.62	0.52	- 0.10		
WR+ (30)	A	87	0.68	0.56	- 0.12	- 0.08	lethal/ sublethal
	B		0.61	0.57	- 0.04		
	C		0.52	0.45	- 0.07		

were found clinging to the top of the beakers, refusing to burrow into the soil and defying their normal photophobic response to avoid exposure to JHDP soil. As the concentration of JHDP in the soil mixture decreased, so did the lethal and sublethal effects. On the basis of these results, the JHDP soil was found to be significantly toxic to both plants and earthworms.

Soils from the white phosphorus pits were generally nontoxic to plants ($\text{SER} \geq 70\%$). In the earthworm toxicity screening, all soils produced an $\text{ESR} \geq 93\%$. Only JWPPA soil may have had some small toxic effect since 1 of 15 earthworms died, and the average weight difference (-0.05 g) was greater than the WR- controls. No definitive tests were conducted because the JWPPA soil was nontoxic to plants and had minimal, if any, toxicity to earthworms.

The two soil samples tested from the riot control pits did have a negative impact on the SER. In phytotoxicity screening tests, JBT1W had a 40% SER, and JBTMA had a 0% SER. Although the SERs from subsequent definitive tests ranged from 55 to 83% (relatively low), the results of the definitive phytotoxicity study of JBT1W soil showed no significant ($p > 0.2763$) difference in plant heights or dry weights ($p > 0.2727$). Results of earthworm toxicity screening of JBT1W soil showed no lethal effects on earthworms; however, JBT1W may have had a sublethal effect on them since it produced a small weight loss during the study. The overall effect was considered minor, and no definitive earthworm test was conducted. On the basis of the definitive phytotoxicity and earthworm screening tests, only minimal toxicity was associated with JBT1W soil. Because the phytotoxicity screening of JBTMA soil produced a 0% SER, a definitive phytotoxicity test was conducted. These results produced SERs ranging from 98 to 100% for JBTMA soil; however, the trend was a decline in plant heights as the amount of JBTMA in the soil mixture increased although this decline was not statistically significant. An analysis of the dry weights indicated a significant (ANOVA: $p < 0.0001$) difference only between the negative control (WR-) and the other treatment levels. The results of definitive phytotoxicity testing indicated that JBTMA soil produced only minor sublethal effects, and none were significant. Results of the earthworm toxicity screening of JBTMA soil produced no lethal/sublethal effects (100% ESR; 0.04 g average weight gain). Therefore, further definitive testing was unnecessary as only minor toxicity was attributed to the JBTMA soil at statistically insignificant levels.

For the soil samples from the adjacent areas, JBPGF had only a 10% SER in the phytotoxicity screening test; however, of the 20 seeds sown, the 2 plants that did emerge survived to Day 14. The JBPCP104 soil had a 30% SER, and of the 20 seeds sown, only 5 of the 6 plants that emerged survived to Day 14. Based on these screening results, definitive phytotoxicity tests were performed on both soils. Definitive phytotoxicity testing of JBPGF soil produced SERs ranging from 48 to 100%. The ANOVA of the plant heights was significantly ($p < 0.0001$) different between WR- and the other treatment levels but not between the JBPGF treatment levels. The ANOVA of the dry weights indicated no significant ($p > 0.05$) difference between treatment levels. WR- had the largest average plant height but the smallest average dry weight; whereas, plants at the 100% level had the smallest average plant height and the largest average dry weight. No significant phytotoxicity is attributable to JBPGF soil. It had no effect on earthworms according to the earthworm screening test (ESR was 100% weight gain). These results coupled with the results from the phytotoxicity tests indicated that this soil is not toxic to either plants or earthworms.

Definitive phytotoxicity testing of JBPCP104 soil produced SERs ranging from 42 to 77%. The ANOVA of plant heights indicated a significant ($p < 0.0001$) negative difference only between WR+ and the other treatment levels. The average dry weights of plants grown in JBPCP104 soil were greater than the dry weights of the controls. These results indicate that JBPCP104 soil has neither lethal nor significant sublethal effects on lettuce seeds. The earthworm toxicity screening test of JBPCP104 soil indicated potential lethal (80% ESR) and sublethal effects (average weight loss of -0.13 g) on earthworms. Therefore, a definitive study was conducted to determine the concentration at which lethal/sublethal effects begin. However, results of the definitive earthworm test of JBPCP104 indicated no lethal effects. The average weight differences indicated that as the amount of JBPCP104 soil in the mixture increased, the weight loss increased; but, there were no significant negative differences among treatment levels. The T-Test was significantly ($p < 0.0091$) different between the 25% level and all other treatment levels except the 50% level. Earthworms in the 25 and 50% levels were the only ones to gain weight during the study; all other groups lost weight. However, the weight losses for these levels (75 and 100%) were not significantly different from both control groups. The trend was toward increasing weight loss as the concentration of JBPCP104 soil in the mixture increased. Because there was no significant difference between the WR-control group and the 75 and 100% levels, the results can not be verified. Since earthworms at the 100% level lost the most weight, this soil has the potential to produce minor sublethal effects.

Therefore, the results of phytotoxicity testing indicates that soil from this location produces no lethal/sublethal effects on either seed germination or early seedling growth. But it does have the potential to produce negative effects on earthworms at the 100% treatment level. It is recommended that additional earthworm toxicity testing be conducted on JBPCP104 soil to confirm the results of this study.

5. CONCLUSIONS

The results of all tests performed on J-Field soils to date (i.e., phytotoxicity screening tests, phytotoxicity definitive tests, earthworm screening tests, and definitive earthworm tests), are summarized in Table 19. Of the 18 soils tested from J-Field, 7 were nontoxic to the species of plants and earthworms used in these studies. Six other soils were placed in the "minimal" category for overall toxicity; 3 soils exhibited moderate toxicity. One soil (JHDP) exhibited high toxicity to earthworms and was given an overall toxicity classification of "high." The JBPMB soil was given an "unknown" based on the results of the screening tests. Definitive studies should be conducted on this soil to determine its overall toxicity.

This study has shown the importance of using multiple bioassays for examining National Priority List sites. Multiple bioassay screening and definitive tests use different trophic levels that can help determine areas within a site where additional testing should be conducted. For example, a particular contaminant that may produce a toxic response in one organism may not cause toxicity in another. Therefore, testing with bioassay containing different trophic levels, as performed in this study, helped identify both toxic sites and those sites or areas within sites that need further investigation.

Table 19. Summary of Results of Toxicity Testing of J-Field Soils

Soil ID	Plant Toxicity Screen Defin.	Earthworm Toxicity Screen	Earthworm Toxicity Defin.	Overall Toxicity	Interpretation of Results
<u>Toxic Burning Pits</u>					
JBP2C	none	NA	sublethal	NA	minimal effects on earthworms
JBP1C	none	NA	sublethal	NA	minimal effects on earthworms
JBPMB	lethal	NA	I/s	NA	significant: plant; moderate: earthworms
JBP2C	none	NA	none	NA	no significant toxic effects
JBPMA	none	NA	none	NA	no significant toxic effects
JBP2A	none	NA	sublethal	NA	minimal effects on earthworms
JBP2B	none	NA	I/s	moderate	moderate effects on earthworms
JBP2C	NA	NA	I/s	moderate	moderate effects on earthworms
JHDP	lethal	none	I/s	high	significant toxicity to earthworms
JBP2P	NA	none	sublethal	NA	minimal effects on earthworms
<u>White Phosphorus Pits</u>					
JWP1E	none	NA	none	NA	no significant toxic effects
JWPPB	none	NA	none	NA	no significant toxic effects
JWP2C	none	NA	none	NA	no significant toxic effects
JWPPA	none	NA	I/s	moderate	moderate effects on earthworms
<u>Riot Control Pits</u>					
JBT1W	I/s	none	sublethal	NA	minimal effects on earthworms
JBTMA	lethal	none	none	NA	no significant toxic effects
<u>Adjacent Areas</u>					
JBP2G	I/s	none	none	NA	no significant toxic effects
JBP2P104	I/s	none	I/s	sublethal	minimal effects on earthworms

* I/s = lethal/sublethal

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APPENDIX A

PLANT DATA

Heights of Lettuce Plants Grown in J-Field Soils

Table A-1. Heights of Lettuce Plants on Day 14, Grown in J-Field Soils -
Phytotoxicity Screening of Soils from Toxic Burning Pits

Plant No.	Soil ID									
	JBP2C	JBP1C	JBPMB	JBPPC	JBPMA	JBPPA	JBPPB	JHDP	WR-	WR+
	Plant Heights (mm)									
1.	10	6	0	7	11	10	14	0	10	3
2.	8	10	0	9	8	10	10	0	6	1
3.	5	8	0	10	6	8	10	0	8	2
4.	10	8	0	10	4	8	14	0	6	0
5.	9	6	0	9	10	9	6	0	7	0
6.	7	8	0	15	9	8	11	0	8	0
7.	9	6	0	10	10	10	9	0	9	0
8.	9	6	0	12	11	8	8	0	9	0
9.	9	8	0	13	10	9	15	0	10	0
10.	9	6	0	13	10	8	12	0	8	0

Table A-2. Heights of Lettuce Plants on Day 14, Grown in JHDP Soil at Different Concentrations (JHDP Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR+
Replicate	Plant No.	Plant Heights (mm)					
A	1.	13	7	5	7	7	4
	2.	10	9	11	7	5	4
	3.	10	6	8	6	7	4
	4.	10	5	12	3	4	0
	5.	6	4	8	7	8	0
	6.	8	9	5	5	7	0
	7.	5	9	0	3	5	0
	8.	7	7	0	4	5	0
	9.	0	0	0	5	6	0
	10.	0	0	0	0	0	0
B	1.	8	8	9	8	4	6
	2.	8	7	9	7	7	0
	3.	10	13	8	6	4	0
	4.	6	9	7	3	5	0
	5.	5	12	6	3	4	0
	6.	7	11	8	2	6	0
	7.	9	8	6	0	5	0
	8.	5	7	8	0	6	0
	9.	8	9	7	0	5	0
	10.	0	0	0	0	0	0
C	1.	12	6	6	6	4	5
	2.	7	5	6	7	5	5
	3.	8	6	7	5	6	10
	4.	7	9	9	6	4	7
	5.	8	6	6	3	5	5
	6.	6	6	7	5	4	0
	7.	0	7	7	4	3	0
	8.	0	8	10	4	5	0
	9.	0	7	8	4	9	0
	10.	0	7	9	5	1	0
Mean:		7.96	7.67	7.68	5.00	5.37	5.56
Std Dev :		2.14	2.09	1.75	1.66	1.42	1.94

Appendix A

Table A-3. Heights of Lettuce Plants on Day 15, Grown in JBPNP Soil at Different Concentrations (JBPNP Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR+
Replicate	Plant No.	Plant Heights (mm)					
A	1.	12	12	15	13	16	4
	2.	16	19	16	17	15	5
	3.	13	14	17	13	17	3
	4.	15	13	17	15	13	3
	5.	8	17	16	14	15	1
	6.	14	12	14	13	16	4
	7.	17	15	16	13	14	3
	8.	20	15	17	11	15	0
	9.	15	12	16	13	12	0
	10.	13	15	16	12	10	0
B	1.	16	15	11	16	16	3
	2.	14	14	14	12	17	3
	3.	18	16	10	17	14	4
	4.	15	13	14	16	12	5
	5.	20	12	13	18	12	0
	6.	11	18	13	15	11	0
	7.	14	20	16	13	14	0
	8.	17	13	18	15	15	0
	9.	16	12	15	11	16	0
	10.	16	18	14	16	16	0
C	1.	13	13	12	11	11	3
	2.	15	13	14	14	14	2
	3.	11	12	13	15	12	0
	4.	15	16	14	13	10	0
	5.	11	14	13	13	13	0
	6.	13	13	12	12	14	0
	7.	11	12	13	15	15	0
	8.	16	14	13	13	13	0
	9.	17	17	14	13	14	0
	10.	0	12	11	13	12	0
Mean:		14.55	14.30	14.23	13.73	13.80	3.36
Std Dev :		2.75	2.25	1.99	1.76	1.99	1.08

Appendix A

Table A-4. Heights of Lettuce Plants on Day 14, Grown in Soils from White Phosphorus Pits - Phytotoxicity Screening

Plant No.	Soil ID					
	JWP1E	JWPPB	JWP2C	JWPPA	WR-	WR+
	Plant Heights (mm)					
1.	10	10	11	12	10	3
2.	10	12	12	13	6	1
3.	10	10	8	12	8	2
4.	12	9	11	15	6	0
5.	11	10	10	15	7	0
6.	7	10	10	11	8	0
7.	8	15	10	12	9	0
8.	8	15	10	11	9	0
9.	9	13	10	9	10	0
10.	10	14	10	6	8	0

Table A-5. Heights of Lettuce Plants on Day 14, Grown in Soils from Riot Control Pits - Phytotoxicity Screening

Plant No.	Soil ID			
	JBT1W	JBTMA	WR-	WR+
	Plant Heights (mm)			
1.	10	0	10	3
2.	10	0	6	1
3.	10	0	8	2
4.	12	0	6	0
5.	10	0	7	0
6.	11	0	8	0
7.	15	0	9	0
8.	12	0	9	0
9.	0	0	10	0
10.	0	0	8	0

Table A-6. Heights of Lettuce Plants on Day 14, Grown in JBT1W Soil at Different Concentrations (JBT1W Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR +
Replicate	Plant No.	Plant Heights (mm)					
A	1.	11	9	14	9	9	0
	2.	13	8	11	9	10	0
	3.	14	8	9	7	8	0
	4.	10	9	12	6	11	0
	5.	10	8	10	6	9	0
	6.	8	9	10	7	10	0
	7.	9	10	10	11	11	0
	8.	10	9	9	9	12	0
	9.	14	8	8	7	11	0
	10.	14	9	10	6	9	0
B	1.	7	12	11	17	10	0
	2.	6	7	12	11	10	0
	3.	6	10	9	11	9	0
	4.	8	8	13	7	11	0
	5.	9	10	11	13	13	0
	6.	11	11	10	17	10	0
	7.	10	12	10	0	8	0
	8.	9	10	8	0	12	0
	9.	7	11	10	0	13	0
	10.	6	8	12	0	14	0
C	1.	9	13	10	10	13	0
	2.	7	14	9	9	7	0
	3.	9	10	8	14	11	0
	4.	12	12	5	12	9	0
	5.	12	9	7	12	10	0
	6.	10	15	8	13	8	0
	7.	11	15	8	15	0	0
	8.	9	16	9	14	0	0
	9.	8	12	8	15	0	0
	10.	8	13	8	11	0	0
Mean:		9.57	10.50	9.63	10.69	10.31	0.00
Std Dev :		2.34	2.42	1.88	3.38	1.78	0.00

Appendix A

Table A-7. Heights of Lettuce Plants on Day 14, Grown in JBTMA Soil at Different Concentrations (JBTMA Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR+
Replicate	Plant No.	Plant Heights (mm)					
A	1.	17	19	13	7	11	8
	2.	14	17	11	11	8	4
	3.	15	12	9	9	11	3
	4.	13	15	12	10	7	2
	5.	20	11	14	9	8	3
	6.	17	15	12	12	11	3
	7.	15	16	15	9	9	0
	8.	14	14	11	13	10	0
	9.	15	17	14	9	7	0
	10.	14	12	12	9	8	0
B	1.	14	10	11	13	9	6
	2.	24	9	13	12	8	7
	3.	19	8	14	13	7	6
	4.	13	14	11	12	10	5
	5.	17	12	10	14	11	6
	6.	16	9	12	10	16	5
	7.	17	10	11	14	8	6
	8.	17	13	12	14	10	5
	9.	16	11	14	13	9	4
	10.	17	14	14	12	11	0
C	1.	20	11	17	9	10	3
	2.	19	14	14	11	13	5
	3.	17	15	17	15	7	7
	4.	15	16	13	10	16	7
	5.	16	12	16	12	11	6
	6.	15	11	13	10	16	8
	7.	19	15	11	11	14	7
	8.	13	13	17	12	13	8
	9.	16	17	10	13	17	0
	10.	20	18	17	14	14	0
Mean:		16.47	13.33	13.07	11.40	10.67	5.39
Std Dev :		2.54	2.87	2.24	2.01	2.99	1.80

Appendix A

Table A-8. Heights of Lettuce Plants on Day 14, Grown in Soils from Adjacent Areas - Phytotoxicity Screening

Plant No.	Soil ID			
	JBPCP104	JBPGF	WR-	WR+
	Plant Heights (mm)			
1.	4	5	10	3
2.	4	7	6	1
3.	5	0	8	2
4.	3	0	6	0
5.	7	0	7	0
6.	0	0	8	0
7.	0	0	9	0
8.	0	0	9	0
9.	0	0	10	0
10.	0	0	8	0

Table A-9. Heights of Lettuce Plants on Day 15, Grown in JBPGF Soil at Different Concentrations (JBPGF Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR+
Replicate	Plant No.	Plant Heights (mm)					
A	1.	10	7	9	9	5	0
	2.	8	8	8	8	7	0
	3.	10	7	9	9	6	0
	4.	12	10	10	7	8	0
	5.	9	8	6	8	6	0
	6.	4	9	7	9	5	0
	7.	0	8	8	8	11	0
	8.	0	14	9	9	9	0
	9.	0	8	6	8	4	0
	10.	0	6	6	9	6	0
B	1.	8	5	6	8	9	0
	2.	13	12	5	6	7	0
	3.	8	7	10	8	8	0
	4.	13	6	6	6	7	0
	5.	16	5	10	8	6	0
	6.	17	8	7	7	7	0
	7.	11	5	8	8	6	0
	8.	13	8	7	9	7	0
	9.	7	5	8	8	8	0
	10.	5	7	9	7	7	0
C	1.	7	9	9	7	7	0
	2.	11	5	8	6	8	0
	3.	13	8	9	7	7	0
	4.	15	12	8	11	8	0
	5.	13	9	9	7	6	0
	6.	9	10	8	8	7	0
	7.	13	8	8	7	8	0
	8.	7	7	8	8	7	0
	9.	9	8	8	10	8	0
	10.	6	7	11	9	11	0
Mean:		10.27	7.87	8.00	7.97	7.20	0.00
Std Dev :		3.39	2.18	1.44	1.16	1.54	0.00

Appendix A

Table A-10. Heights of Lettuce Plants on Day 14, Grown in JBPCP104 Soil at Different Concentrations (JBPCP104 Soil/WR Soil)

		Concentrations (%)					
		WR-	25	50	75	100	WR+
Replicate	Plant No.	Plant Heights (mm)					
A	1.	10	11	11	8	7	5
	2.	12	11	9	11	8	4
	3.	9	14	8	15	11	0
	4.	9	8	9	9	8	0
	5.	11	9	9	5	7	0
	6.	10	8	8	8	9	0
	7.	5	7	10	10	11	0
	8.	10	9	12	7	9	0
	9.	0	10	11	5	7	0
	10.	0	8	9	8	9	0
B	1.	22	14	11	9	11	6
	2.	15	9	8	7	7	5
	3.	11	10	10	10	9	0
	4.	9	9	8	12	8	0
	5.	6	15	9	11	6	0
	6.	8	11	13	10	9	0
	7.	8	11	12	9	9	0
	8.	10	9	10	8	10	0
	9.	9	10	15	10	10	0
	10.	0	10	12	9	8	0
C	1.	10	6	12	9	10	7
	2.	12	11	7	4	11	0
	3.	7	7	15	6	12	0
	4.	11	11	8	7	9	0
	5.	12	14	11	7	9	0
	6.	13	10	11	8	10	0
	7.	10	9	10	9	10	0
	8.	8	10	9	9	9	0
	9.	0	9	14	6	9	0
	10.	0	11	10	0	7	0
Mean:		10.28	10.03	10.37	8.59	8.97	5.40
Std Dev :		3.27	2.14	2.09	2.23	1.47	1.14

Appendix A

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APPENDIX B

STATISTICAL DATA-PLANTS

Analysis of Variance (ANOVA) of Heights of Lettuce Plants Grown
in J-Field Soils
and
Student-Newman-Keuls Analysis of Treatment of Lettuce Plant Heights
and Dry Weights Grown in J-Field Soils

Table B-1. ANOVA of Plant Heights of Lettuce Grown in JHDP Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	23	27	25	25	27	9
Mean:	7.96	7.67	7.68	5.00	5.37	5.56
Std Dev	2.14	2.09	1.75	1.66	1.42	1.94
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	653.6176		135			
Error:	436.9150		130		3.3609	
Treatment:	216.7026		5		43.3405	12.90
Significant at p < 0.0001						

Table B-2. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Heights (mm), Grown in JHDP Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
WR-	A	1.7010	6
50	A	1.6268	5
25	A	1.5305	4
WR+	B	1.3942	3
100	B	1.1634	2
75	B		

Table B-3. ANOVA of Plant Dry Weights of Lettuce Grown in JHDP Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	3	3	3	3	3	3
Mean:	0.0069	0.0079	0.0087	0.0067	0.0064	0.0023
Std Dev	0.0012	0.0008	0.0026	0.0019	0.0012	0.0007
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	0.000102		17			
Error:	0.000028		12		0.000002	
Treatment:	0.000074		5		0.000015	6.30
Significant at p < 0.0043						

Table B-4. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Dry Weights (g), Grown in JHDP Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
50	A	0.0042	6
25	A	0.0040	5
WR-	A	0.0037	4
75	A	0.0033	3
100	A	0.0027	2
WR+	B		

Table B-5. ANOVA of Plant Heights of Lettuce Grown in JBPNP Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	29	30	30	30	30	14
Mean:	14.55	14.30	14.23	13.73	13.80	3.36
Std Dev	2.75	2.25	1.99	1.76	1.99	1.08
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	2189.7178		162			
Error:	692.7200		157		4.4122	
Treatment:	1496.9978		5		299.3996	67.86
Significant at p < 0.0001						

Table B-6. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Heights (mm), Grown in JBPNP Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
WR-	A	1.7115	6
25	A	1.6370	5
50	A	1.5404	4
100	A	1.4036	3
75	A	1.1716	2
WR+	B		

Table B-7. ANOVA of Plant Dry Weights of Lettuce Grown in JBPNP Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	3	3	3	3	3	3
Mean:	0.0069	0.0085	0.0098	0.0110	0.0123	0.0007
Std Dev	0.0006	0.0017	0.0012	0.0004	0.0013	0.0006
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	0.0003		17			
Error:	0.0000		12		0.0000	
Treatment:	0.0003		5		0.0000	45.58
Significant at p < 0.0001						

Table B-8. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked From High to Low: Lettuce Plant Dry Weights (g), Grown in JBPNP Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
100	A	0.0029	6
75	AB	0.0027	5
50	BC	0.0026	4
25	CD	0.0023	3
WR-	D	0.0019	2
WR+	E		

Table B-9. ANOVA of Plant Heights of Lettuce Grown in JBT1W Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	30	30	30	26	26	0
Mean:	9.57	10.50	9.63	10.69	10.31	0.00
Std Dev	2.34	2.42	1.88	3.38	1.78	0.00
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	826.9648		141			
Error:	796.9103		137		5.8169	
Treatment:	30.0545		4		7.5136	1.29
Not Significant at p > 0.2763						

Table B-10. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Heights (mm), Grown in JBT1W Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
75	A	1.7737	5
25	A	1.6688	4
100	A	1.5203	3
50	A	1.2687	2
WR-	A		

Table B-11. ANOVA of Plant Dry Weights of Lettuce Grown in JBT1W Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	3	3	3	3	3	0
Mean:	0.0065	0.0080	0.0076	0.0070	0.0059	0.0000
Std Dev	0.0016	0.0006	0.0009	0.0010	0.0015	0.0000
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	0.0000		14			
Error:	0.0000		10		0.0000	
Treatment:	0.0000		4		0.0000	1.51
Not Significant at p > 0.2727						

Table B-12. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Dry Weights (g), Grown in JBT1W Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
25	A	0.0031	5
50	A	0.0029	4
75	A	0.0026	3
WR-	A	0.0021	2
100	A		

Table B-13. ANOVA of Plant Heights of Lettuce Grown in JBTMA Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	30	30	30	30	30	23
Mean:	16.47	13.33	13.07	11.40	10.67	5.39
Std Dev	2.54	2.87	2.24	2.01	2.99	1.80
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	2773.9075		172			
Error:	1019.3449		167		6.1039	
Treatment:	1754.5626		5		350.9125	57.49
Significant at p < 0.0001						

Table B-14. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Heights (mm), Grown in JBTMA Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
WR-	A	1.8852	6
25	B	1.8033	5
50	B	1.6969	4
75	C	1.5463	3
100	C	1.2909	2
WR+	D		

Table B-15. ANOVA of Plant Dry Weights of Lettuce Grown in JBTMA Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	3	3	3	3	3	3
Mean:	0.0073	0.0049	0.0045	0.0045	0.0045	0.0013
Std Dev	0.0008	0.0003	0.0004	0.0006	0.0009	0.0003
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	0.00006		17			
Error:	0.00000		12		0.00000	
Treatment:	0.00005		5		0.00001	32.68
Significant at p < 0.0001						

Table B-16. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Dry Weights (g), Grown in JBTMA Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
WR-	A	0.0016	6
25	B	0.0015	5
100	B	0.0014	4
50	B	0.0013	3
75	B	0.0010	2
WR+	C		

Table B-17. ANOVA of Plant Heights of Lettuce Grown in JBPGF Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	26	30	30	30	30	0
Mean:	10.27	7.87	8.00	7.97	7.20	0.00
Std Dev	3.39	2.18	1.44	1.16	1.54	0.00
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	739.8356		145			
Error:	592.3487		141		4.2011	
Treatment:	147.4869		4		36.8717	8.78
Significant at p < 0.0001						

Table B-18. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked From High to Low: Lettuce Plant Heights (mm), Grown in JBPGF Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
WR-	A	1.4848	5
50	B	1.3970	4
75	B	1.2727	3
25	B	1.0622	2
100	B		

Table B-19. ANOVA of Plant Dry Weights of Lettuce Grown in JBPGF Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR+
N:	3	3	3	3	3	0
Mean:	0.0033	0.0053	0.0046	0.0038	0.0060	0.0000
Std Dev	0.0015	0.0017	0.0004	0.0002	0.0004	0.0000

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Square	F Value
Total:	0.00003	14		
Error:	0.00001	10	0.00000	
Treatment:	0.00001	4	0.00000	3.09
Not Significant at $p > 0.05$				

Table B-20. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Dry Weights (g), Grown in JBPGF Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
100	A	0.0029	5
25	A	0.0027	4
50	A	0.0024	3
75	A	0.0019	2
WR-	A		

Table B-21. ANOVA of Plant Heights of Lettuce Grown in JBPCP104 Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	25	30	30	29	30	5
Mean:	10.28	10.03	10.37	8.59	8.97	5.40
Std Dev	3.27	2.14	2.09	2.23	1.47	1.14
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	887.2349		148			
Error:	724.1745		143		5.0642	
Treatment:	163.0604		5		32.6121	6.44
Significant at p < 0.0001						

Table B-22. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Heights (mm), Grown in JBPCP104 Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
50	A	2.2968	6
WR-	A	2.1967	5
25	A	2.0669	4
100	A	1.8831	3
75	A	1.5716	2
WR+	B		

Table B-23. ANOVA of Plant Dry Weights of Lettuce Grown in JBPCP104 Soil

	Concentrations (%)					
	WR-	25	50	75	100	WR +
N:	3	3	3	3	3	3
Mean:	0.0043	0.0097	0.0107	0.0103	0.0083	0.0012
Std Dev	0.0006	0.0006	0.0021	0.0012	0.0006	0.0002
Source of Variation	Sum of Squares		Degrees of Freedom		Mean Square	F Value
Total:	0.00023		17			
Error:	0.00001		12		0.00000	
Treatment:	0.00022		5		0.00004	39.01
Significant at p < 0.0001						

Table B-24. Student-Newman-Keuls Analysis of All Treatments, Pairwise, and Ranked from High to Low: Lettuce Plant Dry Weights (g), Grown in JBPCP104 Soil (Means with the same letter are not significantly different; $p > 0.05$)

Lettuce Plants			
Soil	Grouping	Range	Means
50	A	0.0029	6
75	A	0.0028	5
25	A	0.0026	4
100	A	0.0023	3
WR-	B	0.0019	2
WR+	C		

APPENDIX C

SEED EMERGENCE RATES OF LETTUCE PLANTED IN J-FIELD SOILS

Table C-1. Emergence Rates (%) of Lettuce Seeds Planted in Soils from the Toxic Burning Pits; % (Number Emerging/Number Planted)

<u>Screening Tests (20 Seeds/Soil Sample)</u>									
Soil :	JBP2C	JBP1C	JBPMB	JBPPC	JBPMA	JBPPA	JBPPB	JHDP	WR- WR +
% :	100	100	100	75	75	75	60	0	65 25

<u>Definitive Tests (60 Seeds/Treatment)</u>			
<u>JHDP Soil</u>	<u>%</u>	<u>JBNP Soil</u>	<u>%</u>
WR-	38	WR-	95
25%	53	25%	95
50%	55	50%	82
75%	47	75%	97
100%	75	100%	95
WR +	15	WR +	23

Table C-2. Emergence Rates (%) of Lettuce Seeds Planted in Soils from the White Phosphorus Pits; % (Number Emerging/Number Planted)

<u>Screening Tests (20 Seeds/Soil Sample)</u>					
Soil :	JWP1E	JWPPB	JWP2C	JWPPA	WR- WR +
% :	95	80	70	70	65 25

Table C-3. Emergence Rates (%) of Lettuce Seeds Planted in Soils from the Riot Control Pits; % (Number Emerging/Number Planted)

<u>Screening Tests (20 Seeds/Soil Sample)</u>				
Soil : JBT1W JBTMA WR- WR+				
% :	40	50	65	25
<u>Definitive Tests (60 Seeds/Treatment)</u>				
JBT1W Soil	%	JBTMA Soil	%	
WR-	62	WR-	93	
25%	82	25%	98	
50%	83	50%	100	
75%	62	75%	100	
100%	55	100%	98	
WR+	0	WR+	38	

Table C-4. Emergence Rates (%) of Lettuce Seeds Planted in Soils from the Adjacent Areas; % (Number Emerging/Number Planted)

<u>Screening Tests (20 Seeds/Soil Sample)</u>				
Soil : JBPCP104 JBPGF WR- WR+				
% :	30	10	65	25
<u>Definitive Tests (60 Seeds/Treatment)</u>				
JBPCP104 Soil	%	JBPGF Soil	%	
WR-	42	WR-	48	
25%	77	25%	82	
50%	62	50%	93	
75%	63	75%	100	
100%	63	100%	92	
WR+	10	WR+	0	

APPENDIX D

STATISTICAL DATA-EARTHWORMS

Analysis of Covariance (ANCOVA) of Weight Differences of Earthworms
Raised in J-Field Soils

and

T-Test Analysis Among Adjusted Weight Means (g) of Earthworms
Raised in J-Field Soils

Table D-1. ANCOVA of Weight Differences (g) of Earthworms in JHPD Soils

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Squares	F Value	Significance Level
Soil Site	0.07879	4	0.01970	19.71	0.0001

Table D-2. T-Test Analysis of All Treatments: Final Weights (g) of Earthworms in JHDP Soil

Treatment	100%	25%	50%	75%	WR-
<u>p values</u>					
100%					
25%	0.0001				
50%	0.0020	0.0131			
75%	0.0059	0.0050	0.5828		
WR-	0.0001	0.8972	0.0076	0.0026	

Table D-3. ANCOVA of Weight Differences (g) of Earthworms
in JBPCP104 Soils

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Squares	F Value	Significance Level
Soil Site	0.02579	5	0.00516	8.51	0.0016

Table D-4. T-Test Analysis of All Treatments: Final Weights (g)
of Earthworms in JBPCP104 Soil

Treatment	WR-	WR+	25%	50%	75%	100%
<u>p values</u>						
WR-						
WR+	0.0562					
25%	0.0091	0.0002				
50%	0.0644	0.0016	0.3077			
75%	0.4223	0.2228	0.0019	0.0133		
100%	0.1359	0.6090	0.0006	0.0043	0.4640	

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